

AMENDMENTS TO THE SPECIFICATION

Please replace the paragraph on page 3, lines 30-35, with the following paragraph, marked-up to show changes made.

[7] The monitor protein according to [1] wherein the luminescent protein is luciferase derived from anyone selected from the group consisting of *Cypridina noctiluca* *Vargula hilgendorfi*, *Acanthephya purpurea*, luminescent insects (firefly, headlight beetle, etc.), luminescent *Pyrocystis lunula*, luminescent earthworm, *Latia neritoides*, *Renilla*, and *Aequorea victoria* (aequorin).

Please replace the paragraph on page 4, lines 7-12, with the following paragraph, marked-up to show changes made.

[9] The monitor protein according to [2] characterized in that the luminescent protein in the property variable region is a light-emitting enzyme from secretory *Cypridina noctiluca* *Vargula hilgendorfi* and the fluorescent protein in the property variable region is a yellow fluorescent protein from a mutant derived from luminescent *Aequorea victoria*.

Please replace the paragraph on page 6, line 30 to page 7, line 10, with the following paragraph, marked-up to show changes made.

In a preferred embodiment, in the monitor protein, the luminescent protein and the fluorescent protein are arranged at both termini of the property variable region, respectively. In the monitor protein, either one may be present at the N-terminus, but it is more preferable that the luminescent protein and the fluorescent protein are present in this order from the N-terminus. In the monitor protein having the luminescent protein and the fluorescent protein, when the processing does not occur, the monitor protein emits yellow-green light by the energy transfer of blue light emitted from the luminescent protein (secretory *Cypridina noctiluca* *Vargula hilgendorfi*, light-emitting enzyme) to the fluorescent protein mutant (EYFP). When the processing has occurred, the energy transfer is not ~~effected~~ affected and only the blue light is emitted from the monitor protein. The processing can be analyzed using this color change of the emitted light in the property variable region as the indicator.

Please replace the paragraph on page 10, line 24 to page 11, line 1, with the following paragraph, marked-up to show changes made.

As the luminescent protein, luciferases derived from various luminescent organisms such as *Cypridina noctiluca**Vargula hilgendorfi*, *Acanthephya purpurea*, luminescent insects (firefly, headlight beetle, etc.), luminescent earthworm, *Latia neritoides*, *Renilla* and *Aequorea victoria* (aequorin) are exemplified. For example, the *Cypridina noctiluca**Vargula hilgendorfi* luciferase is a secretory type, and is preferable because the monitor protein also becomes secretory when this is used. When the secretory monitor protein is used, a level of the processing which occurs in living cells can be evaluated without disrupting the cells. In the case of non-secretory luciferase such as luciferase derived from *Renilla*, it can be utilized as a secretory luminescent protein by introducing a secretory protein into the N-terminal side.

Please replace Table 2 between lines 13 and 15, with the following paragraph, marked-up to show changes made.

Table 2

| Biological luminescent protein | Fluorescent protein |
|---|-----------------------------------|
| <i>Cypridina noctiluca</i> <u><i>Vargula hilgendorfi</i></u> luciferase | GFP, YFP, BFP, CFP, DsRED, RFP. |
| Firefly luciferase | DsRED, Phycocyanin, Phycoerythrin |
| Luminescent <i>Pyrocystis lunula</i> luciferase | GFP, YFP, BFP, CFP, DsRED, RFP |
| Headlight beetle luciferase | DsRED |
| <i>Renilla</i> luciferase | GFP, YFP, BFP, CFP, DsRED, RFP |
| Aequorin | GFP, YFP, BFP, CFP, DsRED, RFP. |

Please replace the paragraph on page 13, line 27 to page 14, line 5, with the following paragraph, marked-up to show changes made.

The protein of the present invention can be obtained by incorporating the gene of the present invention described later into the expression vector and expressing it in appropriate host cells. As the expression vector, for example, pBT-VL-mp-YFP (VL, mp and YFP indicate *Cypridina noctiluca* *Vargula hilgendorfi* luciferase, a monitor peptide and the yellow fluorescent protein, respectively) and the like can be used. The host cells include eukaryotic cells such as mammalian cells and yeast cells, and prokaryotic cells such as *Escherichia coli*, *Bacillus subtilis*, algal and fungal cells, and any of them may be used. As the preferable host cell, mammalian cultured cell COS7 cell line (in this system, it is important to go through protein synthesis and protein modification processes of a mammalian system, i.e., this process will be monitored) and the like can be used.

Please replace the paragraph on page 17, line 77 to page 18, line 15, with the following paragraph, marked-up to show changes made.

NG108-15 cells derived from nervous cells were transfected with gene vectors containing three monitor proteins, Vluc-NST/Noc-EYFP, mut-Vluc-NST/Noc-EYFP and del-Vluc-NST/Noc-EYFP using a commercially available transfection reagent. Proteins secreted in culture media were analyzed by a Western blotting method (Fig. 3). Vluc-EYFP which had no insertion sequence was used as a control. An *anti-Cypridina noctiluca* *anti-Vargula hilgendorfi* luciferase antibody (ant-Vluc) recognizes a light-emitting enzyme at the N-terminal side of the monitor protein, and four types of the monitor proteins have a molecular weight of 95 kDa. An anti-green fluorescent protein antibody (anti-GFP) recognizes the fluorescent protein at the C-terminal side of the monitor protein, and four monitor proteins showed the molecular weight of 95 kDa as a main protein. However, in the monitor protein of Vluc-NST/Noc-EYFP, a minor band at 27 kDa compared to the band at 95 kDa was observed. A processing enzyme was expressed in NG108-15 cells derived from the nervous cells, and cleaved the monitor protein, the antibody recognized EYFP at the C-terminal side after the cleavage and consequently this band was produced. In the mutated or deleted, two mut-Vluc-NST/Noc-EYFP and del-Vluc-NST/Noc-EYFP, this 27 kDa band was not detected and it has been demonstrated that the

processing enzyme can strictly recognize the inserted amino acid sequence. But, a 68 kDa band recognized by anti-GFP seems to be artificial. In order to demonstrate that the cleavage which occurs in the monitor protein of Vluc-NST/Noc-EYFP is attributed to either PC1 or PC2 which is a representative processing enzyme which cleaves insulin and enkephalin, PC1 or PC2 was forcibly expressed in the cells into which the monitor protein had been transfected. As a result, it has been demonstrated that PC1 remarkably increased the 27 kDa band compared to the 95 kDa band whereas PC2 scarcely changed. Consequently, it has been demonstrated for the first time that the processing between nocistatin and nociceptin (NST/Noc) occurs due to PC1.